

PRODUCTION OF ALL MALE FINGERLINGS THROUGH FEEDING OF ANDROGENIC STEROID (MESTEROLONE) IN THE DIET FED AS FIRST FOOD TO TILAPIA *Oreochromis niloticus*

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ABSTRACT

Non-conventional hormone, Androgenic Steroid (mesterolone) was prepared and incorporated (using ethanol) at various concentration of 0mg, 30mg, 40mg, 50mg, 60mg each in 0.2kg of fishmeal. The six levels were replicated 3 times in plastic tanks. The fish meal incorporated with hormone (mesterolone) were fed as first food to 7-day old fry of *Oreochromis niloticus* for 28 days after which the tilapia fry were fed on ordinary fishmeal for another 8 weeks. At the 9th and 12th weeks and after feeding with the hormone, the number of survivals and the weights gained were recorded. After the 12th week, the males and females were separated using hand-magnifying lens. The economic viability of all male production using non-conventional hormone (mesterolone) was calculated. Hormone fed to *O. niloticus* was found to have no significant effect ($P > 0.05$) on the survival and the mean weight gain of the *O. niloticus*. But the hormone fed had great influence on the female: male ratio i.e. treatment difference was highly significant ($P < 0.01$). Tilapia fry fed with 40mg/0.2kg feed was found to have the largest percentage of males 97% while those fed with 30mg/0.2kg, 50mg/0.2kg, 60mg/0.2kg and 70mg/0.2kg had 51%, 91%, 86%, 89%, and 80% males respectively.

INTRODUCTION

Tilapia constitutes one of the most commonly cultivated fish species in the world. Although their natural distribution is confined to Africa and Palestine, different species have been transferred to different continents. Since tilapias are cultivated worldwide, this is an indication of its acceptability hence; this work highlights the economic potential of Tilapia. Tilapia has risen not only from contributing significantly to protein production in Africa and the Far East; but it has become established as a food in Latin America (Balarin and Hatton, 1982).

Nigeria was observed as the largest consumer of fish in Africa (FAO, 1980) where the domestic consumption of fish was estimated to be more than one million metric tones. With the rapid increase in the population of Nigeria the percent consumption of fish increased over the years. This resulted in heavy importation of fish by the Nigerian Government. In order to reduce to the barest minimum, importation of fish in Nigeria there is every need to develop our local fish species (Tilapia) that is cheaper and affordable compared to other fish species since it has been adjudged to have great potentials within our local environment.

With all the potentials of Tilapia, excessive reproduction with its resultant problems render tilapia unsuitable for producing high grade fish for consumption (Narcel, 1970). Its production for food has long been hindered by the precocious maturity, and uncontrolled reproduction (Balarin 1982) and (Wohlfarth and Hulata, 1981). These difficulties introduce a setback to commercial production of Tilapia.

Various attempts have been made over the years to overcome the problem such as the all-male reproduction, manual sexing, hybridization, hormonal sex reversal, predator control, genetic control of maturation towards a later stage, generation of infertile fry, (Balarin, 1982). Among all these techniques the elimination of female sex altogether has been favored (Popma and Green, 1990).

Hormonal sex reversal is widely recognized as having significant advantage (Guerrero, 1987 and MC Andrew 1993). The orally active preparation of methyl testosterone, which is conventionally used for sex reversal, is no longer available in the UK and Nigerian market. Methyl testosterone like other 17- alpha alkyl derivatives of testosterone could sometime cause dose-related but reversible cholestatic jaundice (British Medical Association and the Royal Pharmaceutical Society of Great Britain, 1990). The cost of importing this conventional hormone from United States of America is high. Therefore, in order to reduce the reliance upon the conventional hormones, which are unavailable and/ or expensive, and to evade its

associated health risks, a locally available, cheaper and non health risk hormone must be researched.

Mesterolone (proprietary name Mestoranum: Proviron 17 - Hydroxyl alpha methyl androstan 3 one) is not a 17- alpha alkyl derivative and is less toxic to the liver (Narcel, 1970). This study is therefore designed to investigate the potential of mesterolone in producing all male fingerlings of Tilapia (*Oreochromis niloticus*) at varying degree of inclusion in the diet fed as first food to Tilapia.

MATERIALS AND METHODS

The brood fish of *O. niloticus* were collected from the Ministry of Agriculture and Natural Resources, Oyo State, Nigeria and stocked inside concrete tank (5 x 2 x 1) m³ at the back of the Department of Wildlife and Fisheries Management building. The fish were kept under observation until they spawn naturally.

Collection of fry was carried out carefully by using fry scoop net; fry collected were released into well-aerated water and transferred to the laboratory. Measurements of some fry caught were taken and close observation revealed that they have not absorbed their yolk completely. Eighteen fry of *O. niloticus* were allotted per tank with three replicates per treatment. Each tank contained 11 liters of aerated water, which was completely changed every other day.

Mesterolone was obtained from a Pharmaceutical shop in Ibadan and fishmeal from a livestock feed mill also in Ibadan. Different levels of hormone 70mg, 60mg, 50mg, 40mg and 30mg active ingredient were differently dissolved in 50ml of ethanol. The solution of each level was used to moisten the feed (0.2kg fish meal containing 72% crude protein) to form diets I, II, III, IV, V and VI.

The control diet was moistened with ethanol but without the hormone. The diets were spread under room temperature to allow the ethanol to evaporate, and kept in dried sealed containers. The feeding experiment lasted for 28 days after which feeding of the fry on ordinary fishmeal (72% crude protein) commenced for eight weeks. Weighing was done at the 9th and 12th week and hand lens was used in sexing the fish after 12 weeks of culture.

RESULT AND DISCUSSION

The average weights of fish in the 9th and 12th week are recorded in Tables 1(a) and 1 (b) respectively

Table 1.0 (a) Average Weight of Fish in the 9th Week (g)

Treatment (mg)	Replicate 1	Replicate 2	Replicate 3	Mean
0	0.53	0.62	0.63	0.59
30	0.55	0.43	0.68	0.55
40	0.61	0.91	0.55	0.69
50	0.62	0.69	0.8	0.69
60	0.57	0.61	0.87	0.68
70	0.67	0.87	0.84	0.79

Table 1.0 (b) Average Weight of Fish in the 12th Week (g)

Treatment (mg)	Replicate I	Replicate 2	Replicate 3	Mean
0	0.73	0.56	0.56	0.72
30	0.73	0.59	0.59	0.67
40	0.76	1.16	1.16	0.89
50	0.80	0.83	0.83	0.74
60	0.76	0.76	0.76	0.86
70	0.80	1.11	0.80	0.94

The number of survivals after the 4th and 12th weeks are recorded in Table 2.0 (a) and 2.0 (b) respectively
 Table 2.0 (a) Number of survivors after the 4th week.0

Treatment (mg)	Replicate I	Replicate 2	Replicate 3	Mean
0	15	15	15	15.00
30	16	14	15	15.00
40	16	11	16	14.33
50	08	17	13	12.67
60	11	17	13	13.67
70	14	10	14	12.67

Table 2.0 (b) Number of survivors after the 12th week

Treatment (mg)	Replicate I	Replicate 2	Replicate 3	Mean
0	13	14	14	13.67
30	16	14	15	15.00
40	15	11	15	13.67
50	08	17	11	12.00
60	10	17	11	12.67
70	13	10	12	11.67

The highest mortality was recorded in the fish fed with diet VI (70mg hormone) and the least mean mortality was recorded in the fish fed diet II.

Sexing of Experimental fish

The experimental fish were separated into the male and female. Table 3.0 (a) shows the ratio of males to females among the experimental fish.

Table 3.0(a): The ratio of male to female in each replicate

Treatment (mg)	Replicate 1		Replicate 2		Replicate 3	
	Male	Female	Male	Female	Male	Female
0	06	07	08	06	07	07
30	15	01	13	01	13	02
40	15	-	10	01	15	-
50	06	02	16	01	09	02
60	10	-	14	03	10	01
70	10	03	10	-	08	04

Table 3.0 (b): The value of female: Male ratio

Treatment (mg)	Replicate 1	Replicate 2	Replicate 3	Mean ratio of female to male
0	1.17	0.75	1.00	0.97
30	0.07	0.08	0.15	0.10
40	0.00	0.10	0.00	0.03
50	0.33	0.66	0.22	0.20
60	0.00	0.21	0.01	0.10
70	0.30	0.00	0.05	0.27

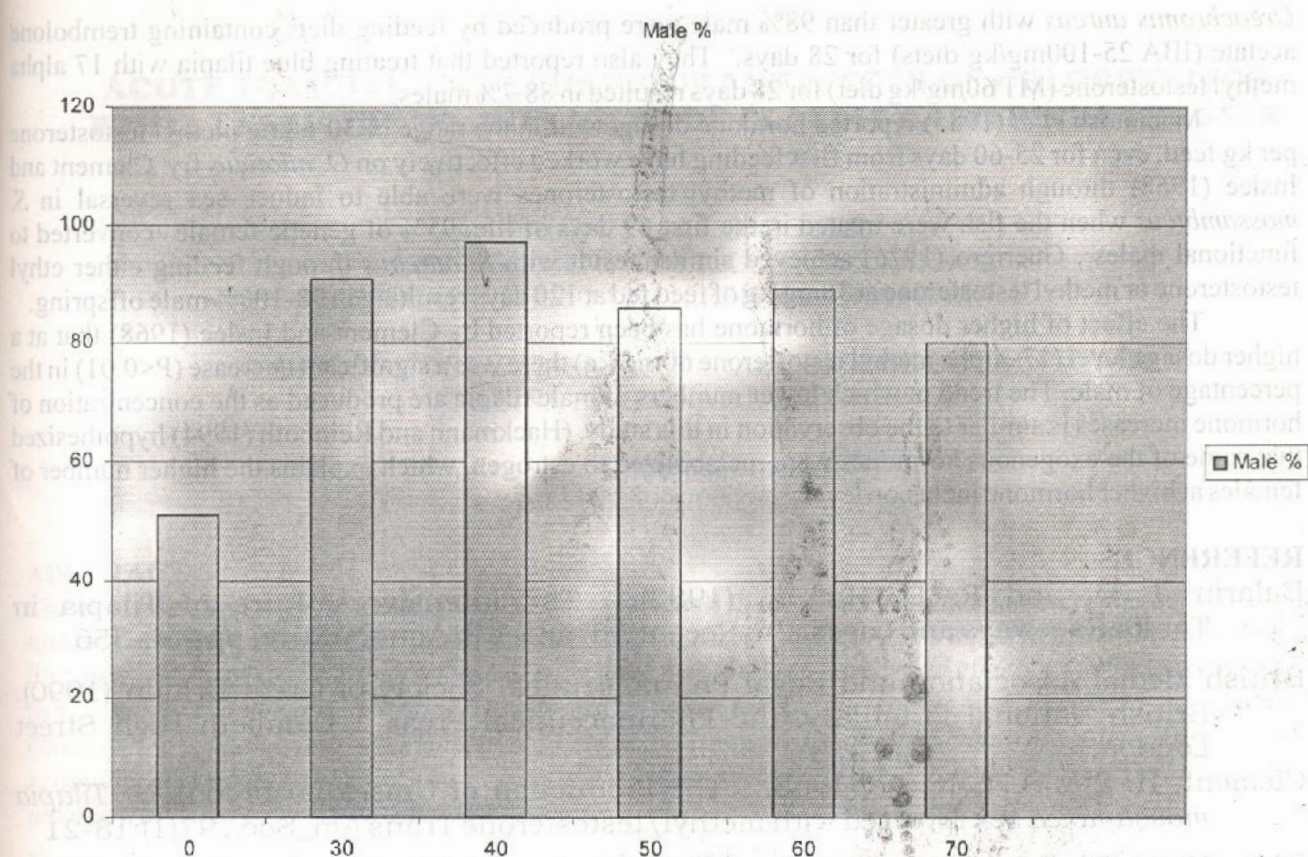


Fig. 1: The Percentage of males obtained

Statistical analysis of variance shows that there was no significant difference ($P > 0.05$) in the mean weight of tilapia fed with different levels of hormone (Androgenic steroid). However the highest mean weight gained was obtained from Tilapia fed with feed containing 70mg of the hormone incorporated while the fish with the least mean weight gain had 30mg hormone inclusion.

SEX RATIO AND RATE OF MALE PRODUCTION

Analysis of variance revealed that there was significant difference ($P < 0.05$) in the sex ratio of fish produced as a result of various levels of hormone incorporated into their feed. Least significant difference further reveals that the treatment level that had the highest female to male ratio (0.97) was fish treated with 0mg of hormone-incorporated feeds. The treatment difference was highly significant at ($P < 0.01$).

However this was followed by 0.27, 0.20, 0.1, 0.1, 0.03 female to male ratio in feed that contain 70, 50, 60, 30, 40 hormones incorporated feed respectively. Fish with 40mg hormone incorporated feed had the lowest female and highest number of male tilapia produced. This is vividly shown in Fig. 1.

ECONOMIC VIABILITY OF ALL MALE TILAPIA

Total weight of feed (hormone incorporated with 40mg/0.2kg) fed to a tank in 28 days was 16.8g. Total cost of feed to the fish in 28 weeks was N7.81k and the average number of male got from tank fed with 40mg/0.2kg feed was thirteen, hence the cost of feed used to produced each male tilapia was 60 kobo.

DISCUSSION

The study revealed that the highest percentage of male (97%) fish production was obtained in the experimental fish (*Oreochromis niloticus*) fed with feed containing 40mg hormone. The best male percentage (89% and 91%) male production was obtained in the feeds incorporated with hormones ranging between 30mg and 60mg. Towards the extreme ends of the range, the percentage of the male produced reduced. The control (0mg) produced the lowest percentage of male and the highest percentage of female. As the level of hormone included increases there is reduction in the percentage of male fish produced. It can therefore be inferred that the higher the levels of hormone included in the feed (after 40mg), the lower the percentage of male fish produced. Galvez *et al* (1996) reported that pooled population of the tilapia

Oreochromis aureus with greater than 98% male were produced by feeding diets containing trembolone acetate (IBA 25-100mg/kg diets) for 28 days. They also reported that treating blue tilapia with 17 alpha methyl testosterone (MT 60mg/kg diet) for 28 days resulted in 88.7% males.

Macintosh *et al* (1985) reported hormone dosage within the range of 30-60mg methyl testosterone per kg feed, even for 25-60 days from first feeding have worked effectively on *O. niloticus* fry. Clement and Inslee (1968) through administration of methyl testosterone were able to induce sex reversal in *S. mossambicus* when the fish were treated in the first 69 days of life, 95% of genetic female converted to functional males. Guerrero (1976) achieved similar result with *S. aureus* through feeding either ethyl testosterone or methyl testosterone at 30mg/kg of feed fed at 120 days resulting in 98-100% male offspring.

The effect of higher dosage of hormone has been reported by Clement and Inslee (1968) that at a higher dosage level (17- alpha methyl testosterone 60mg/kg) there was a significant decrease ($P < 0.01$) in the percentage of male. The trend in which lower numbers of male tilapia are produced as the concentration of hormone increases is similar to the observation in this study. (Hackmann and Reinboth (1994) hypothesized that some of the exogenous hormones were metabolized to estrogen, which explains the higher number of females at higher hormone inclusion levels.

REFERENCES

- Balarin J. D. and R. D. Hatton, (1982). The intensive culture of Tilapia in Tanks, Raceways and cages. In: Recent Advances in Aquaculture, pp 266-356
- British Medical Association and Royal Pharmaceutical Society of Great Britain (1990). British National formulary, the Pharmaceutical Press 1 Cambeth High Street London.
- Clement, H. P & S. T. Inslee (1968): The Production of Unisexual broods by *Tilapia mossambica* sex reversed with methyl/testosterone Trans Am. Soc., 97(1):18-21
- FAO, (1980) Report on the adhoc consultation on Aquaculture Research 3 9 May 1980 FAO/Fisheries Report No 238
- Galvez J.I.; Morrison J.B; Phelps R.P. (1996) Efficacy of trembolone acetate in sex inversion of the blue tilapia *Oreochromis aureus*: In: Journal of the World Aquaculture Society 1996, 27:4, 483 486
- Guerrero R. D. (1987) Tilapia farming in the Philippine Technology and livelihood Resources Centre, Manila Philippines.
- Guerrero, R. D. (1976): Culture of Male Tilapia *mossambica* produced through artificial sex reversal. Proc. FAO Tech Conf. on Agriculture, Kyoto Japan. FAO AQ/Conf./76/E.15. 3p
- Huet M. (1980) Textbook of Fish Culture Breeding and Cultivation of fish Edition Ch. De Wyngaret, Brussels Fish in News (Books) Ltd.
- Macintosh D. J.; Varghese, T.J. and G.P.S Rao (1985) Hormonal sex reversal of wild spawned tilapia in India J. Fish Biol., 26: 87-94
- Mc Andrew B. J. (1993). Sex Control in Tilapia. In: Recent Advances in Aquaculture IV (eds. J.F. Muir and R. J. Roberts). Pp 87 98. Blackwell Science Oxford.
- Narcel (1970): Effect of body's natural testosterone production @ [www. the festival.net/feed?id=194-46k](http://www.the-festival.net/feed?id=194-46k)
- Popma. T. J. and Green, B. W (1990) Aquaculture Production. Manual sex reversal or Tilapia in Earthen ponds Research and Development series No 35 Auburn University, Alabama, USA.
- Hackmann, E and Reinboth, R., (1974). Delimitation of the critical stage of hormone influenced sex differentiation in *Hemihaplochromis multicolour* (Hilendorf) (Cichlidae). Gen. Comp. Endocrinol. 22: 42-53.
- Wohlfarth, G. W. and Hulata, G. I. (1981). Applied Genetics of Tilapia, International Centre for living Aquatic Resource Management, Manilla, The Philippines. ICLARM Studies and Review 6:26pp.